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Accumulation and Retention of Dietary ^{14}C-DDT by Atlantic Menhaden[1]

STANLEY M. WARLEN, DOUGLAS A. WOLFE,
CURTIS W. LEWIS, AND DAVID R. COLBY
National Marine Fisheries Service, Atlantic Estuarine Fisheries Center
Beaufort, North Carolina 28516

ABSTRACT

The accumulation and retention of dietary ^{14}C-DDT by young-of-the-year menhaden, and the effect of ^{14}C-DDT exposure on growth of menhaden were studied. A simple mathematical model was developed to predict DDT flux in a natural menhaden population.

Uptake of ^{14}C-DDT was dose dependent and a function of exposure time. Prediction equations summarize the ^{14}C-DDT uptake by menhaden for each of three exposure levels. Menhaden assimilated and retained 17 to 27 percent of the cumulative dosages of ^{14}C-DDT. The biological half-life of ^{14}C-DDT in menhaden was estimated to be 428, 64, and 137 days, respectively, for the low (0.58 ppb), medium (9.0 ppb), and high (93 ppb) dose groups. Exposure to dietary ^{14}C-DDT at these concentrations did not produce any effect on growth of menhaden nor did starvation significantly affect their retention of ^{14}C-DDT.

The linear DDT flux model for menhaden was:

$$FD_f a = (\gamma + \lambda)\ (D_m),$$

where F is the daily feeding rate, D_f is the DDT concentration in the food, a is the fraction of the ingested DDT that is assimilated, γ is the daily fractional DDT accumulation rate, λ is the daily fractional DDT turnover rate, and D_m is the DDT concentration in menhaden. The model was tested with the experimental data and also used as a basis for comparison of the experimental data with previous field observations.

Relatively little is known regarding the uptake and elimination of DDT by estuarine fishes. Research on the dietary uptake and subsequent loss of organochlorine insecticides by fish has been limited to freshwater or anadromous species. Buhler et al. (1969) determined the oral toxicity, and the amounts of DDT accumulated by juvenile coho (*Oncorhynchus kisutch*) and chinook (*O. tshawytscha*) salmon. Grzenda et al. (1970) studied the uptake, metabolism, and elimination of ^{14}C-DDT fed to goldfish, and Macek et al. (1970) did a similar study on rainbow trout. Studies of long-term dietary uptake and subsequent body burden loss of DDT by estuarine fish are not available.

We initiated this study to provide estimates of the rates of accumulation and retention of DDT by a commercially and ecologically important fish, the Atlantic menhaden, *Brevoortia tyrannus*, during its larval and juvenile stages, and to test the effect of DDT exposure on growth of menhaden. The uptake rate from food and the biological half-life of ^{14}C-DDT at several exposures are described for young-of-the-year menhaden fed several different concentrations of ^{14}C-DDT. Also, we used these data and data from a concurrent field study (Warlen 1974) to develop a mathematical model to predict DDT flux in a natural menhaden population.

METHODS

Atlantic menhaden used in this study were collected on March 15, 1972, with a channel net (Lewis et al. 1970) near Beaufort, North Carolina. In the laboratory, more than 3,000 of the larval fish were placed in shallow tabletop tanks equipped with running seawater. The fish were acclimated to laboratory conditions and fed *Artemia* nauplii, and as they grew, increasing amounts of size-00 Purina Trout Chow.[2] After one week, 300 fish were carefully transferred to each of four experimental tanks where they were acclimated and fed a diet of pesticide-free Trout Chow until the experiment began on April 6. Several fish that

[1] Based on part of a dissertation submitted by the senior author to the Department of Zoology, North Carolina State University, in partial fulfillment of the requirements for the degree of Doctor of Philosophy.

[2] Reference to trade names does not imply endorsement by the National Marine Fisheries Service, NOAA.

died during this acclimation were replaced with fish from the stock kept in the table-top tanks.

The experiments were conducted in a part of the open seawater system described by Hettler et al. (1971). The 600-liter capacity fiberglass tanks had smooth inside walls painted black and floors painted white. Tank tops with screens prevented foreign matter from entering the tanks. Drainage standpipes, covered with screen to prevent fish loss, were adjusted so each tank contained approximately 125 liters of water. Unfiltered water, maintained at a salinity of 15‰ and a temperature of at least 20 C, flowed into each tank from an overhead reservoir at a rate of 0.5 to 1.0 liter/minute.

Experimental Diets

Size-00 Trout Chow was sieved and particles between 125 and 800 μ were collected and then cleaned of detectable organochlorine insecticide residues by using a petroleum ether Soxhlet extraction. A new diet was reconstituted by thoroughly mixing the following constituents on a weight basis in petroleum ether: 88% cleaned, sized Chow, 8% corn oil cleaned according to Stober and Payne (1966), 2% vitamin fortification mixture (ICN Nutritional Biochemicals, Cleveland, Ohio), and 2% vegetable lecithin as a fat binder. The petroleum ether was then evaporated. A stock batch of ^{14}C-DDT labeled food was made by using the same technique, and in addition, 10 parts per million (ppm) ringlabeled ^{14}C-*p,p'*-DDT (specific activity 23.9 mCi/mmol or 149.4 dpm/ngDDT) of 99% purity was added in the petroleum ether. We serially diluted the labeled stock with clean reconstituted food to obtain diets theoretically containing 1, 10, and 100 parts per billion (ppb) DDT. Analyses showed that the food actually contained 0.58, 9.0, and 93 ppb ^{14}C-DDT, respectively. The ^{14}C labeled *p,p'*-DDT was obtained from Amersham/Searle Corp., Arlington Heights, Illinois.

Experiments

Fish (mean total length of 32 mm and mean dry weight of 14 mg) were first fed the ^{14}C-DDT treated food on April 6. Fish in tanks I–IV received diets containing 93 ppb DDT, 9.0 ppb DDT, 0.58 ppb DDT, and no DDT (control), respectively. Fish were fed a daily ration equal to 2–3% of their previous week's mean wet body weight. The ratio of dry fish weight to wet fish weight was 0.22. Food was slowly sprinkled on the water, which allowed the fish to eat virtually all of it before any sank to the bottom of the tank. Tanks were checked daily, the rate of inflowing water was adjusted, and any dead fish were removed. Tanks were cleaned twice weekly to prevent buildup of feces, silt, and the attachment of organisms to the tank walls and floor, and several times during the experiment we used large bulb pipettes to withdraw samples of detritus that had accumulated on the tank floors. The samples of detritus were placed in glass jars, the water decanted, and the material was lyophilized for analysis of ^{14}C-DDT. The total particulate and dissolved ^{14}C-DDT in water was determined from water samples taken periodically from the tanks.

The three groups of experimental fish received labeled food for 48 days (uptake period) and then were fed unlabeled food until the experiment ended on day 157 (retention period). Fish fed the low, medium, and high dose diets received 25, 392, and 4,054 ng of ^{14}C-DDT, respectively, during the 48-day exposure period.

Menhaden from each tank were sampled on days 1, 2, 4, 8, 16, 24, 32, 40, 48, 49, 51, 53, 55, 57, 61, 65, 69, and then every eight days through day 157. Individual samples were composites of 10 fish each through day 48 and 4 to 6 fish from day 49 to day 157. All fish were measured to the nearest mm total length and the composite samples were weighed to the nearest 10 mg wet weight and 0.1 mg dry weight. Growth rates for fish of each tank were obtained from linear regressions of dry weight on time.

To study the effect of starvation on the retention of DDT, we moved 30 fish from tank I where they had been exposed to 93 ppb DDT in food, to another tank where they were starved for 33 days beginning on day 109. The starved fish were sampled at the same times as the fish remaining in tank I.

Analysis of ^{14}C-DDT

Samples of whole larval and juvenile menhaden were frozen and were then lyophilized.

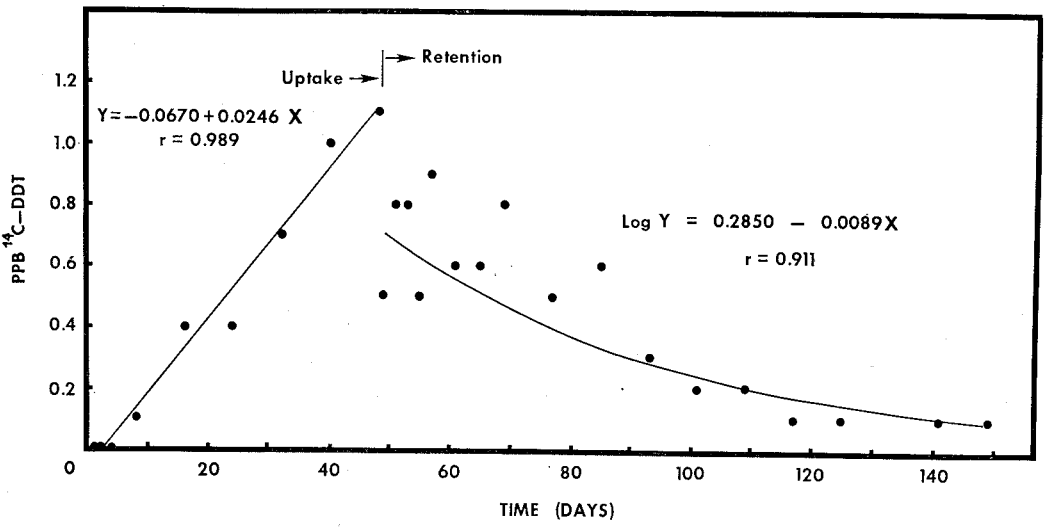

FIGURE 1.—*Accumulation and retention of ^{14}C-DDT by Atlantic menhaden—low dose, 0.58 ppb. Regressions are of ppb DDT on time. Each data point for uptake is a composite of 10 fish and for retention is 4–6 fish.*

The dried samples were extracted with three successive 5-ml quantities of petroleum ether in a size-22 Duall tissue grinder. The centrifuged extracts were placed on a size-22 Chromoflex column packed with Florisil (Floridin Company, Berkeley Springs, West Virginia) topped with sodium sulfate and eluted with 20 ml of 1% ethyl ether in petroleum ether, which elutes DDD, DDE, and DDT residues (Alfred J. Wilson, Jr., personal communication). The eluate was then evaporated to 5 ml, quantitatively transferred to a scintillation vial, and re-evaporated to 1 ml before adding 15 ml of scintillation fluid. Each 100-ml water sample was extracted with two successive 10-ml portions of petroleum ether. The limit of detection of ^{14}C-DDT in water samples was $<1 \times 10^{-3}$ ppb. The extracts for a sample were combined, dried over anhydrous sodium sulfate, evaporated to 5 ml, and transferred to a liquid scintillation vial as described above. The scintillation fluid contained 4 g PPO (2,5-diphenyloxazole) and 100 mg POPOP (*p*-Bis [2-(5-phenyloxazolyl)]-benzene) per liter of reagent grade toluene.

The extraction efficiencies for ^{14}C-DDT from water, fish, and detritus were 95, 80+, and 80+%, respectively. Samples were counted to a precision of ±2% (2 σ), in a Beckman LS-200B liquid scintillation counter. Counting efficiency was always ⩾94% and background counts were automatically subtracted during the counting. Concentration of ^{14}C-DDT, in ppb, in samples was obtained by dividing the counts per minute per g of dry fish by the specific activity of the stock ^{14}C-DDT. All ^{14}C in samples was assumed to be ^{14}C-DDT despite the possibility that small amounts of ^{14}C-DDD and ^{14}C-DDE could also have been present.

DDT (and DDD, DDE) concentrations in menhaden prior to the experiment were analyzed by gas-liquid chromatography according to the methods of Warlen (1974).

In the several instances in which regression models were fit to experimental data, the "extra sums of squares" principle (Draper and Smith 1966) was applied in selecting a specific model from among several possible models.

RESULTS AND DISCUSSION

^{14}C-DDT Uptake

The net accumulations of ^{14}C-DDT by fish at three exposure levels showed no evidence of any significant curvilinearity (Figs. 1–3). The simple linear models fit the data well with correlation coefficients above 0.97 for the three groups. Macek and Korn (1970) also found that accumulation of DDT and metabolites in

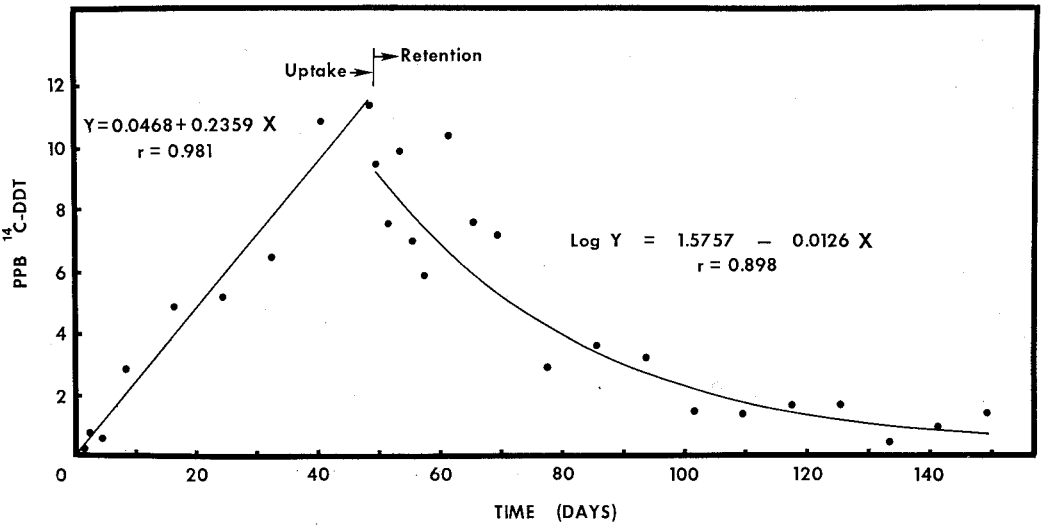

FIGURE 2.—*Accumulation and retention of ^{14}C-DDT by Atlantic menhaden—medium dose, 9.0 ppb. Regressions are of ppb DDT on time. Each data point for uptake is a composite of 10 fish and for retention is 4–6 fish.*

brook trout increased linearly during the first 60 days of dietary exposure to DDT.

An equilibrium between ^{14}C-DDT uptake and loss at any of the experimental doses was apparently not reached since there was no evidence of an upper asymptote on any of the uptake curves. Slopes of the three uptake regression equations were significantly different from zero and from each other. Also, the uptake of ^{14}C-DDT appears directly proportional to dose in the linear phase because the regression coefficients, which (slopes) estimate the daily increase of DDT concentration in the experimental menhaden, differed by about a factor of 10 between the low and medium, and the medium and high exposure

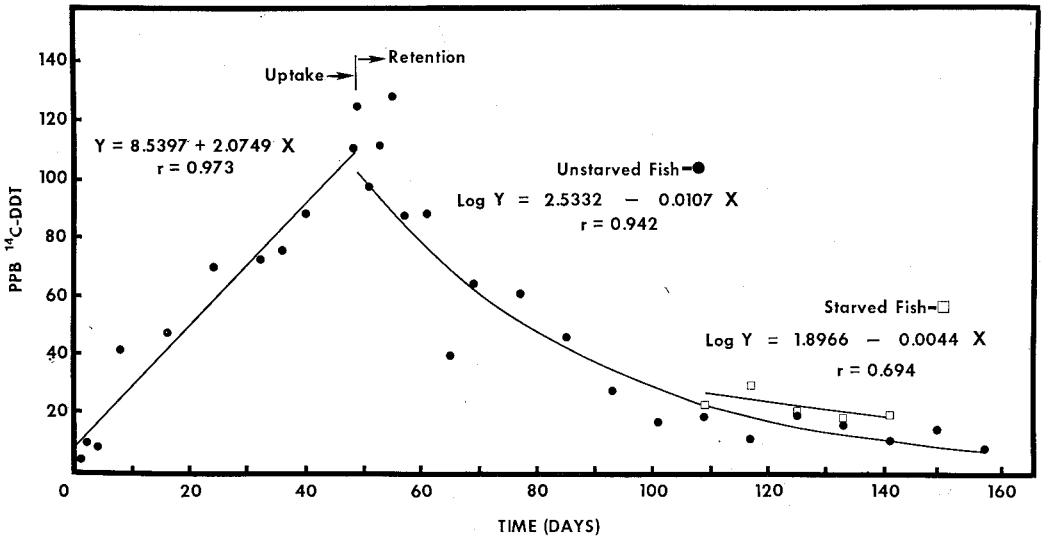

FIGURE 3.—*Accumulation and retention of ^{14}C-DDT by Atlantic menhaden—dose, 93 ppb. Regressions are of ppb DDT on time. Each data point for uptake is a composite of 10 fish and for retention is 4–6 fish.*

TABLE 1.—*Growth rate of menhaden exposed to three levels of DDT in diet. Regression in $\log_{10}$ transformed mean dry weight (mg) on time, log* $Y = a + b X$.

Dietary level	Sample size	Initial size-log (a)	Growth rate (b)	Standard error of b (s_b)	Coefficient of determination (r^2)
Control	28	1.1654	0.0087	0.0004	0.956
Low	28	1.1757	0.0082	0.0004	0.949
Medium	28	1.1449	0.0078	0.0003	0.964
High	28	1.1773	0.0085	0.0003	0.971
Composite	112	1.1658	0.0083	0.0002	0.951

fish. Macek et al. (1970) found that accumulation of both DDT and dieldrin by rainbow trout was also dose dependent. Robinson (1967) stated that for vertebrates the concentration of an organochlorine insecticide in a particular tissue is a function of the daily intake.

Accumulation of DDT by fish in the laboratory did not differ statistically from accumulation in wild fish. Accumulation data for the high dose fish were put on a body burden basis (pg/fish) by multiplying the concentration of ^{14}C-DDT by the predicted dry weight—the latter from Table 1. Body burden was then regressed on time resulting in the following prediction equation:

$$\log Y = 2.1437 + 0.0352\,X. \qquad (1)$$

This can be compared with the equation,

$$\log Y = 2.4827 + 0.0150\,X, \qquad (2)$$

for body burden of environmentally acquired total DDT (DDD + DDE + DDT) on time for larval menhaden from the Newport River estuary (Warlen 1974). Although the daily $\log_{10}$ accumulation rate of 0.0352 for the high exposure fish was approximately 2.35 times the rate for the wild fish, the rates were not significantly different ($P > 0.05$). Only the regressions for the high exposure fish and the wild fish were compared because the respective dietary DDT concentrations the two groups received were similar, 93 ppb and 113 ppb (mean amount of total DDT in the plankton-detritus available to fish caught in 1971, Warlen 1974).

Data on ^{14}C-DDT accumulation by all three menhaden test groups (low, medium, high dose exposures) were combined for multiple regression analysis and yielded the following prediction equation with a regression coefficient of 0.988:

$$\text{ppb } ^{14}\text{C-DDT} = -0.1706 + 0.0865(\text{dose}) + 0.0208(\text{time} \times \text{dose}). \qquad (3)$$

A reduction in sum of squares partial F test showed that this model was significantly better ($P < 0.01$) than a simpler model without the "dose" term and equally as good as a model with a "time" term added. This regression model allows rapid estimation of the ^{14}C-DDT concentration (ppb) in menhaden given various combinations of dose and exposure time.

^{14}C-DDT Assimilation

The percent of the cumulative amounts of ^{14}C-DDT fed to the experimental fish that was retained during the uptake period is shown in Table 2. The calculations are biased estimates of true ^{14}C-DDT assimilation because they include DDT turnover, and they do not take into account that menhaden consumed only 80–90% of their daily ration. Actual assimilation of ingested DDT could be 20% or more than shown in Table 2. The values in Table 2 are comparable to literature values for other species. Chinook salmon retained 12–25% and coho salmon 38–68% of the DDT that they ingested (Buhler et al. 1969). Another salmonid, the rainbow trout, accumulated 20–24% of the available dietary DDT (Macek et al. 1970).

TABLE 2.—*Percent of cumulative dosages of ^{14}C-DDT retained by juvenile menhaden during the 48-day uptake period.*

Time (days)	Exposure group		
	Low	Medium	High
4	8	4	8
8	12	18	21
16	37	20	19
24	20	15	26
32	34	16	21
40	39	23	18
48	41	24	19
Mean	27	17	19

TABLE 3.—*Concentration of ^{14}C-DDT in detritus from experimental tanks (ppb dry weight basis). ND = non-detectable.*

Time (days)	Exposure groups			
	Control	Low	Medium	High
		Uptake		
40	1.1	3.2	6.1	47.0
48	ND	1.0	1.6	23.3
		Retention		
54	ND	<1.0	<1.0	2.5
68	ND	<1.0	<1.0	3.0

The ^{14}C-DDT in detritus on the tank floors during the uptake phase of the experiment (Table 3) probably was from the uneaten food, food ingested but not assimilated, and excreted products. Samples were taken 24 h after the previous day's feeding; each sample consisted of all detritus (100–400 mg dry weight) that could be collected from the tank floor. The mean organic content of ashed aliquots of day 68 samples was 48%. The detritus probably included uneaten food with an organic content of 90%, a mixture of feces and excretion products with an estimated organic content of less than 40% and suspended material (plankton-detritus) with an organic content of 21% (Warlen 1974). Most of the ^{14}C-DDT was present in the uneaten or unassimilated food fraction because the concentration in detritus dropped appreciably after the fish was placed on a ^{14}C-DDT-free diet beginning day 49. The ^{14}C-DDT in detritus after day 49 was probably that lost by menhaden through excretory processes other than defecation.

No ^{14}C-DDT was detected in 100-ml surface water samples. During the uptake period water from only one of 32 samples showed radioactivity of more than 2 cpm over background. Although 1-liter water samples may have given a better estimate, on the basis of the 100-ml samples we concluded that there were negligible concentrations of ^{14}C-DDT in the water.

^{14}C-DDT Loss

When the fish were placed on diets free of ^{14}C-DDT all three groups showed an exponential reduction in concentration with time (Figs. 1–3). The correlation coefficients for the three groups were greater than 0.89. The difference in the values of the correlation coefficients for the respective uptake and loss models may be due to the difference in size of the composite samples, 10 in each uptake sample vs. five in each loss sample. The regression slopes that estimated the logarithmic rate of apparent residue loss were not significantly different from each other ($P > 0.05$) but were significantly different from zero ($P < 0.01$).

The good fit ($r > 0.89$) of these curves resulted mainly from the dominant effect that growth of the fish had on the change in the ^{14}C-DDT concentration in the fish. The retained ^{14}C-DDT was "diluted" with new tissue. Lake Michigan coho salmon fry whose DDT concentration decreased, excreted almost none of the compound but just diluted it by growth (Wilford et al. 1969). There was also an actual loss of ^{14}C-DDT in each menhaden exposure group, which was described by the regression of total body burden (cpm/fish) on time. The data for the low exposure fish were fitted by a linear regression and the data for the medium and high exposure fish were fitted by exponential regressions. The regression slopes were tested and found to be significantly different from zero ($P < 0.05$), indicating an actual loss of ^{14}C-DDT over time. These regressions of body burden on time were not used in the development of the flux model because their correlation coefficients were relatively low, less than 0.542.

Apparent half-lives calculated from the loss equations in Figures 1 to 3 were 34, 24, and 28 days, respectively, for the low, medium, and high dose fish. When the concentrations are converted to body burdens using the weight regressions summarized in Table 1, these recalculated biological half-lives become 428, 64, and 137 days, respectively. We assumed these calculated biological half-lives were not significantly different, since the slopes of the loss equations (Figs. 1–3) were not significantly different ($P > 0.05$) from each other and there was no significant difference ($P > 0.05$) among the growth rates. Gakstatter and Weiss (1967) found that small bluegills, *Lepomis macrochirus*, and goldfish, *Carassius auratus*, eliminated less than 50% (apparent loss) of their experimentally accumulated DDT in 32 days of recovery. They did not

attempt to estimate the half-life of DDT in those species but for each it would be greater than 32 days. The predicted half-life of total body DDT in growing rainbow trout was 160 days (Macek et al. 1970). In contrast, growing fingerling chinook salmon retained 45% of DDT fed them at the end of a 32-day period on DDT-free food and growing fingerling coho salmon retained 45–84% of DDT after 53 days on a DDT-free diet (Buhler et al. 1969). In goldfish the DDT half-life was approximately 30 days according to Grzenda et al. (1970). They gave no data on the size of fish during their experiments.

Effect of Starvation on Loss

Menhaden that were fed increased their body weight by 87% while the starved menhaden lost 42% of their body weight during the 33-day test. The loss of body weight by the starved fish suggests that menhaden in the experiment were subsisting primarily on the prepared diet and were getting very little food from the incoming water.

Despite the loss of 42% of their body weight during the 33-day test, starved juvenile menhaden maintained a relatively constant ^{14}C-DDT concentration. The regression for the DDT concentration in starved fish (Fig. 3) had a slope that is not significantly different from zero ($P > 0.05$), indicating no significant change in their ^{14}C-DDT concentration. The reason the ^{14}C-DDT concentration did not change significantly was due to the loss in weight (42%) and the corresponding calculated loss in ^{14}C-DDT body burden (cpm/fish) of about 58%. Macek (1968) reported a different situation for starved DDT-treated brook trout which exhibited a decrease in body weight and an apparent increase in the concentration of DDT.

The change in ^{14}C-DDT concentration of the starved fish (Fig. 3) was not significantly different ($P > 0.05$) from those of fish that were fed, and thus, starvation of juvenile menhaden produced no detectable effect on their concentration of ^{14}C-DDT, at least for a period of 33 days. However, the absolute loss of ^{14}C-DDT in fed fish was only about half that of the loss by starved fish.

Effect on Growth

Exposure to dietary levels of up to 100 ppb ^{14}C-DDT for 48 days had no effect on the growth of menhaden (Table 1). Linear regressions of the log transformed dry weights on time produced excellent fits for the data as indicated by the high r^2 values. The logarithmic growth rates were all significantly different from zero ($P < 0.01$) but were not significantly different from each other ($P > 0.05$).

The effects of DDT on growth have been examined for several other species of fish. Exposure to several dietary levels (0.2–1.0 ppm) of DDT did not affect the growth of rainbow trout (Macek et al. 1970); however, Macek (1968) suggested that a diet containing 2 mg DDT/kg may have stimulated growth of brook trout.

DDT Flux Model

From the foregoing information on DDT accumulation and retention we can formulate a simple linear model that will approximate the flux of DDT in young-of-the-year Atlantic menhaden. Data from the high dose group of menhaden were used because the dietary DDT exposure of these fish was nearest that estimated for wild menhaden from plankton-detritus, 113 ppb (Warlen 1974). Most simply, the model is of the same form used by Small et al. (1973) and Cross et al. (1975) to describe the flux of Zn in euphausiids and Mn, Fe, Cu, and Zn in estuarine fish. Our proposed model is: Assimilation rate = retention rate + turnover rate or,

$$FD_f a = (\gamma + \lambda)\ (D_m), \qquad (4)$$

where F is the daily feeding rate in mg dry weight food/mg dry weight of fish, D_f is the DDT concentration (ppb) in the food as pg DDT/mg dry weight food, a is the fraction of the ingested DDT that is assimilated, γ is the fraction of DDT in the fish added each day (fractional accumulation rate), λ is the fraction of DDT turned over per day, and D_m is the DDT concentration (ppb) in menhaden as pg DDT/mg dry weight. It is assumed in the model that all DDT in menhaden is obtained via their diet.

Fractional Accumulation Rate (γ)

The body burden of DDT in laboratory-held menhaden increased during the 48-day uptake experiment according to Equation 1. This regression equation expressed exponentially as

$$\text{pg}^{14}\text{C-DDT} = 139.3376e^{0.08107t}, \quad (5)$$

was the product of the exponential expression of the growth equation for high exposure fish from Table 1,

$$\text{mg body weight} = 15.0446e^{0.01958t}, \quad (6)$$

and an equation fit to the rectified (log transformed) concentration values for accumulation of ^{14}C-DDT by the high dose menhaden,

$$\text{ppb}^{14}\text{C-DDT} = 9.2616e^{0.06149t}. \quad (7)$$

The latter equation, though different from the model in Figure 3 and with a lower correlation coefficient ($r = 0.746$), was used so that Equation 5 would be in an exponential form.

Differentiating Equation 5 gives $11.3003 \cdot e^{0.08107t}$ which when divided by the same exponential equation (5) yields a quotient of 0.08107 which is defined as the daily fractional accumulation rate. This rate was calculated with data from menhaden between 15 and 450 mg dry weight.

Fractional Turnover Rate (λ)

Fractional turnover rate of DDT in menhaden is estimated from the ^{14}C-DDT loss data (Fig. 3) where the loss equation can be expressed exponentially as

$$\text{ppb}^{14}\text{C-DDT} = 341.5162e^{-0.02464t}. \quad (8)$$

Since the reduction of DDT concentration in growing fish is a combination of actual loss and a "dilution" of DDT by new tissue, a correction for growth must be applied to put the loss on a body burden basis. This is effected by multiplying Equation 8 by the exponential growth equation for high dose fish, i.e., mg body weight $= 15.0e^{0.01958t}$ to yield:

$$\text{pg}^{14}\text{C-DDT} = 5137.9746e^{-0.00506t}. \quad (9)$$

The absolute value of the exponential coefficient, 0.00506, is the daily fractional turnover rate. The reciprocal of the turnover coefficient, 198, is the biological turnover time in days, which multiplied by the ln of 2, gives the biological half-life of 137 days.

DDT Concentration in Menhaden (D_m)

The total concentration of DDT at any time in the experimental menhaden is the sum of the portion remaining of the initial unlabeled DDT present in the fish at the beginning of the experiment and the net gain of ^{14}C-DDT during the uptake period.

A gas chromatographic analysis of the menhaden at day 0 indicated that they contained 19 ppb DDD, 47 ppb DDE, and 48 ppb DDT. Unlabeled DDD and DDE, although present in the fish, are assumed not to affect the loss of labeled or unlabeled DDT. The initial DDT concentration times the dry body weight at day 0 (15.0 mg) gives the body burden of unlabeled DDT. It is assumed for the model that no additional unlabeled DDT was accumulated during the experiment and that the unlabeled DDT had a daily fractional turnover rate identical to that of ^{14}C-DDT, i.e., 0.00506. The exponential equation for the loss of body burden of unlabeled DDT was then

$$\text{pg DDT} = 48\,(15.0)\,e^{-0.00506t}, \quad (10)$$

which when divided by the exponential growth equation (6) gave the loss in concentration of unlabeled DDT as

$$\text{ppb DDT} = 48e^{-0.02464t}. \quad (11)$$

Net gain of ^{14}C-DDT concentration for the high dose fish through the uptake period was best related to body weight by the following linear regression ($r = 0.906$):

$$\text{ppb}^{14}\text{C-DDT} = -23.8980 + 3.3480\,X. \quad (12)$$

With the exponential growth equation substituted for X the product is $-23.8980 + 50.3693 \cdot e^{0.01958t}$. This expression when added to the unlabeled DDT loss expression (11) gives a value for the D_m term in the model as:

$$D_m = 48e^{-0.02464t} + (-23.8980 + 50.3693e^{0.01958t}). \quad (13)$$

Although not used in the model, the prediction equation (3) also provided a means of estimating D_m (^{14}C-DDT only) for experimental fish using "dose" as well as "time."

Feeding Rate (F), *DDT Concentration in Food* (D_f), *and Fraction DDT Assimilated* (a)

Experimental menhaden were fed at an effective daily feeding rate (F) of 0.16 mg/mg of dry fish weight, based on a constant ration of 3% of their previous week's wet weight and assuming that they consumed 90% of the food offered them. Analyses of the dry formulated diet showed that the actual ^{14}C-DDT concentration (D_f) was 93 ppb (pg/mg). The minimum fraction of DDT that was assimilated (a) by menhaden was estimated during the experiment (Table 2). The mean fraction of DDT assimilated during the experiment was 0.21.

Application of the Model

The model may be used to estimate any one of the constant terms (F, D_f, a, γ, or λ) from the remaining values and from the variable function of D_m. For example, if all terms except (a) are substituted into the model (Equation 4), including the above regression (13) for D_m, and solved at 30 and 50 days (times for which estimates of γ are relevant), the calculated assimilation fraction would be 0.519 and 0.719, respectively, for ^{14}C-DDT from the prepared diet. Calculation of (a) for any time greater than 50 days gives unrealistically high values. A sensitivity test showed that the parameter (λ) with our least confident estimate could be halved or doubled and still not affect the calculation of (a) by more than 5%.

These estimated assimilation fractions are comparable with those found by Buhler et al. (1969) and Macek et al. (1970). The DDT assimilation fraction in menhaden may increase as the fish transform from larvae to the juvenile stage, as indicated by the model. During and after this transformation the gut lengthens drastically (June and Carlson 1971). The alimentary tract length is about 14 times longer in a 60 mm fork length juvenile than in a 20 mm fork length larval menhaden. This fact alone could account for a greater assimilation fraction in juvenile menhaden than in larval menhaden. The assimilation fraction of DDT is probably also a function of temperature since temperature affects the time required for passage of food through the gut (Barrington 1957).

Following the suggestion of Robinson (1967), the model may also be used as a basis for comparison of our experimental data with previous field observations. The combined concentration of DDT, DDE, and DDD (ΣDDT) in wild menhaden was described by the following linear regression equation (Warlen 1974):

$$\text{ppb } \Sigma\text{DDT} = 120.4679 + 0.0478 \text{ (mg dry weight)}. \quad (14)$$

Substituting the growth equation obtained for wild menhaden (mg dry body weight = $6.3149e^{0.027718t}$) for dry weight in the latter regression gives the D_m for wild menhaden:

$$D_m = 120.4679 + 0.3019e^{0.027718t}. \quad (15)$$

An estimate of γ for wild young-of-the-year menhaden is available (Warlen 1974) from the regression of ΣDDT body burden on time, $\log Y = 2.6690 + 0.0140 X$. The fractional accumulation rate (γ) for wild fish (0.03224), determined the same way as above, is about 40% of the value for the experimental fish but is based on the accumulation of ΣDDT over a longer time (200 + days).

The rate of ΣDDT assimilation can then be estimated for the wild menhaden using the experimental estimate of the fractional turnover rate (λ) along with the above γ and the DDT concentration in fish (D_m) function:

$$FD_fa = (0.00506 + 0.03224)(120.4679 + 0.3019e^{0.027718t}). \quad (16)$$

Between 0 and 200 days the estimates for FD_fa are between 4.49 and 7.08 ppb ΣDDT per day. Using the predicted value of FD_fa for day 150, 5.16, and possible ranges of 0.05 to 0.20 for F and 0.20 to 0.80 for a, D_f must be within the limits of 32 and 516 ppb DDT for wild menhaden. This estimated range includes the mean ΣDDT concentration for plankton-detritus (113.0 ppb) suggested by Warlen (1974). The general compatibility in the model between the estimates of feeding rates and assimilation efficiencies and the DDT content of plankton-detritus suggests that the plankton-detritus actually was the dietary source of DDT for the wild fish.

In its present form the model cannot be used easily to predict the concentration of DDT in young-of-the-year menhaden (D_m) at any time (t) given any DDT concentration in their food (D_f). This estimation is impossible because in the model DDT concentration (D_m) is not an explicit function of time (t). From the values discussed in this paper for F, D_f, a, γ, and λ, however, one can generate D_m through time by successive iteration of the model on a day-by-day basis, given only an initial value for D_m at a specified time.

Even though the model is limited to young-of-the-year menhaden and the estimates of the parameters could be refined with more observations, it appears to describe accurately the flow of DDT through menhaden. Also, since menhaden are a dominant herbivorous fish of North Carolina estuaries, the model helps describe the flow of DDT through this ecosystem and interpret the ecological consequences of the use of organochlorine insecticides.

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